Research Proposal Form of the Thesis Entitled

"Evaluation of psychological stress level in partially edentulous patients of Ahmedabad and Gandhinagar district restored with fixed dental prosthesis"

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INTRODUCTION

The cornerstones of a healthy lifestyle include a balanced diet, regular exercise, and enough sleep. Maintaining a healthy lifestyle lowers your risk of illness that maintains you active and strong. As stated by the WHO, leading a healthy lifestyle allows individuals to appreciate many parts of your life. It is a style of life which reduces the risk of developing a major illness or passing too soon. It's not enough to be healthy to stay free from illness or disease. It also has to do with one's social, mental, and physical health. "Oral disorders plays an important roll in routine life of all people, regardless of their location, nation, ethnicity, or color".[1] The effects of tooth loss have been discussed in several contexts. Although partial or complete edentulism may not pose a life-threatening hazard, it has significant implications for the affected person and the community in terms of functional and social constraints as well as the use of public resources.[1]

Replacement of missing teeth is necessary to restore patients' entire physical, mental, and social well-being as tooth loss causes morphological or operational changes to the oral tissues that result in resorption, premature eruption of neighbouring tooth, drifting of tooth into edentulous spaces, malocclusion causing TMJ problems, degradation of facial aesthetics, decline in nutrition,trouble speaking and chewing.[2,3] For the restoration of lost teeth, there are several solutions available, including removable partial dentures, removable complete dentures, fixed dental prostheses (FDP), and removable and fixed implant prostheses.[4] In order to replace missing teeth, FDP is the most popular restorative.

Mental stress can rise as a result of dental, systemic, and social diseases.[5,6] Mastication has been linked to parafunctional behaviours such stress-adaptive cheek, lip, and nail biting, according to studies.[7] Because of alterations occurring in the hippocampus and hypothalamus, acquired behaviours have the potential to affect levels of stress indicators.[8]

Numerous markers found in the blood, urine, saliva, cerebrospinal fluid, and gingival crevicular fluid, which are useful to assess psychological stress.[9] Saliva is a non-invasive marker that may be collected with ordinary tools and is simpler to store like other fluids. In the literature, many salivary biomarkers were examined. Salivary alpha-amylase (sAA) is a stronger link and direct correlate of neuronal activity in ANS.[10]

Perceived stress scale (PSS), State Trait Anxiety Inventory state scale, Ardell Wellness Stress Test, visual analogue scale, and social readjustment rating scale are a few of the questionnaires used to assess psychological stress. These questionnaires measure the stress, anxiety, and depression syndromes.[11-13]

There are several studies completed to find the levels of sAA as a biomarker for diagnosing and treating psychological stress.[14,15] The authors are not aware of any studies in prosthodontic research that show that patient's psychologic stress has been reduced by fixed restorations for missing teeth using biomarkers. The study's goal is to compare the levels of sAA in patients who have permanent dental prostheses in place both earlier and later therapy.

STATEMENT OF PROBLEM

It has been observed that the patient with loss of single tooth or multiple teeth has more stress level compared to the patient with fully dentulous arches.

CLINICAL IMPLICATION OF THE STUDY

Previous study has shown that the instant rehabilitation of missing mandibular first molar is crucial for maintaining psychotic health; the implication of this research will help to check the stress level of the patient with multiple missing teeth pre and post restored with fixed dental prosthesis

AIM

- The Motive of the research is to check the levels of psychological stress in partially edentulous person before and after restoration with fixed dental prosthesis.

OBJECTIVE

- To determine the values of salivary alpha amylase in partially edentulous patients pre and post treatment using fixed dental prosthesis.

HYPOTHESIS

Proposed Hypothesis

- The amounts of stress of partially edentulous patients decreases after treating with fixed dental prosthesis.

Null Hypothesis

- There will be no difference in the amounts of stress of partially edentulous patients after treating with fixed dental prosthesis.

REVIEW OF LITERATURE

The review of literature has been described separately under the following headings:

1) Masticatory system, tooth loss and replacement of missing teeth

2) Psychological stress and markers

3) Scales used to measure stress, anxiety, and depression

4) Partially edentulism

5) Salivary stress biomarkers and salivary alpha amylase (sAA)

1) Masticatory system, tooth loss and replacement of missing teeth

Zarb GA. et al (1988)[5]: had observed that while dentists have used a lot of ingenuity to replace missing teeth, it was not possible to mimic the natural environment and attach tooth-root analogues to the host bone. Such an attachment, which involves carefully implanting threaded, unalloyed titanium implants into the bone, is made possible via osseo integration. The situation of the edentulous patient has already changed significantly as a result of the process' confirmed success. The use of Osseo integration as a partial edentulism treatment technique presents fascinating problems for the dentist.

Soboleva U et al (2005)[2]: had used electromyographic recordings to assess the physiology of the masticatory muscles. The association between jaw motions and

muscle activity has been found to be considerably better understood when electromyography and jaw-tracking equipment are used together. Procedures in clinical dentistry have been significantly influenced by understanding of the movements of the mandible during mastication. The primary goal of this study is to provide a succinct explanation of the traditional investigations of the physiology, brain regulation, and function of mastication. Food is broken down during mastication as a prelude to deglutition. This breakdown process is a highly structured combination of digestive and neuromuscular functions. Depending on the type of food being chewed and the individual, the power stroke's duration and forces change. Although it has been proposed that observing masticatory movements may be useful for diagnosing stomatognathic system abnormalities, there isn't enough evidence to support this claim. The amplitude, timing, and length of the chewing cycle of the masticatory muscles movement during chewing vary amongst subjects. It is best to create restorations that are suitable with the patient's functional movement patterns rather than expecting the patient's mastication patterns to adapt to the newly manufactured restorations since tooth guidance has a significant impact on the muscle activity during chewing and swallowing.

Pjetursson BE et al (2008)[4]: have concluded that the conventional abutment tooth-supported Fixed Dental Prosthesis (FDP), implant-supported FDP, or implant-supported Single Crown should be the first treatment option when planning prosthetic rehabilitations. Cantilever tooth-supported FDP, combined tooth-implant-supported FDP, or resinbonded bridges should be considered only as a last resort. The most common consequences with conventional tooth-supported FDP were biological disorders such as cavities and pulp vitality loss. The incidence of biological problems in cantilever FDP was comparable to that of conventional FDP. However, technical problems such as loss of retention and material fractures were more common. The frequency of biological problems like as mucositis and peri-implantitis in implant-supported restorations was comparable for simply implant supported, combined implant-tooth-supported FDP, and implant-supported Single Crown. He also discovered that the most frequently reported technical difficulties were veneer material fractures (ceramic fractures), abutment or screw loosening, and loss of retention. In studies reporting on gold-acrylic reconstructions, veneer material fracturing was more common than metal-ceramic reconstructions. Complications are much more common

in implant-supported reconstructions than in tooth-supported reconstructions. This does not necessarily indicate that the options for corrective action are more inconvenient. Although a range of subjective and objective factors substantially influence the selection of treatment modalities, understanding the survival and complication rates of various reconstructions based on long-term studies surely aids in optimising the decision process.

Gerritsen et al (2010)[3]: have evaluated tooth loss and quality of life outcomes associated to oral health. This correlation seemed unrelated to the included samples' context (such as their cultural background) or the OHRQoL instrument that was employed. The intensity and amount of impairment, however, appear to be context-dependent. He also noted that the severity of OHRQoL impairment is influenced by not only the number of missing teeth, but also their distribution and placement. Given the detrimental effects of tooth loss on OHRQoL, it is crucial to encourage disease prevention strategies when creating health policies for senior citizens. Patients' requests for therapy meant to keep their teeth will probably increase. More research will be needed to determine the effectiveness of preventive measures, and more economic analysis of tooth replacement options is also needed.

2) Psychological stress and markers

Slavicek R. et al (2004)[7]: have said clenching and grinding between the upper and lower teeth is known as bruxism. They have also noted that exceptionally powerful forces can be used during this action for durations longer than those of functional mastication. Numerous dental issues are brought on by these biomechanical pressures, including abfractions, hypersensitivity, periodontal distraction, and temporo-mandibular dysfunction. Psychological stress and emotional strain have long been discussed by bruxism researchers. The idea of stress management is based on the psychological underpinnings of bruxism and the advantages of masticatory muscle action in lessening symptoms of stress, like stomach ulcers. During times of psychic overload, the masticatory organ's clenching and bruxing function serves as an emergency exit. As a result, the masticatory organ's obstruction greatly affects a person's capacity to handle stress. When done properly, bruxism can be recognised as a reliable system prophylactic for all illnesses linked to stress.

Cohen S et al (2007)[6]: have observed the link between psychological stress and illness, particularly in the cases of depression, HIV/AIDS, and cardiovascular disease. Upper respiratory tract infections, asthma, herpes viral infections, autoimmune illnesses, and wound healing are additional conditions where evidence for the impact of stress is beginning to appear. Although it is impossible to prove a direct connection, data from prospective observational studies support the idea that stress plays a significant role in a number of diseases. The findings of these studies, however, are in agreement with natural experiments on the effects of real-world stressor exposure on disease risk, with laboratory studies demonstrating that stress modifies disease-relevant biological processes in humans, and with animal studies looking at stress as a causal factor in disease onset and progression.

Ono Y. et al (2010)[8]: have gone through many researches in animals and humans and shown that mastication keeps the hippocampus, a part of the brain crucial for learning and memory, functioning cognitively. Reduced mastication, an epidemiological risk factor for dementia in humans, impairs spatial memory and worsens the morphology and functionality of hippocampus neurons, especially in older animals. By inhibiting endocrinological and autonomic stress responses, active mastication restores the hippocampus memory process in animals and reduces the sense of stress in humans. By enhancing the activation of the hippocampus and prefrontal cortex, two brain areas crucial for cognitive processing, active mastication enhances sustained cognitive task performance even more. Chronic stress is produced by experimentally-induced abnormal mastication in animals, which in turn inhibits their capacity for spatial learning. The concept that the inhibition of the hypothalamic-pituitary-adrenal (HPA) axis by masticatory stimulation helps to preserve the cognitive functions linked with mastication has been presented in light of the negative connection between mastication and corticosteroids. Research on the mastication-induced improvement of cognitive impairments, its potential connection to the HPA axis, and the neuronal pathways that may be implicated in this process in the hippocampus have all been looked at in this review.

Ma L. et al (2018)[9]: have observed the variations in index responses to stress and noted their significance for creating effective therapies. These individual variations may be

reflected using the three-dimensional Tridimensional Personality Questionnaire (TPQ). In the meanwhile, there is growing curiosity about the connection between psychological stress and non-invasive markers. Our study's goals were to determine whether these three factors were connected to sAA responses and cognitive behaviour assessments. Based on the results of the temperament measurement parameters of the TPQ, all subjects were classified into various groups. The sAA responses were recorded as the index of stress reactions, compared between groups, and presented in conjunction with Stroop patterns and mental arithmetic stress tests. The high reward-dependence (RD) group's sAA levels were found to be considerably higher than those of the low-RD group both after the stress test and during the recovery period. In comparison to the low-NS group, there were considerably more Stroop trials in the high novelty-seeking (NS) group. Individual variations in TPQ personality traits have been seen to be related to various stress response patterns.

3) Scales used to measure stress, anxiety, and depression

Brokelman RB. et al (2012)[13]: have observed in today's health care system that patient happiness is more crucial than ever. Because satisfaction is a multidimensional issue for which there is no gold standard, assessing it is challenging. Utilizing the well-known visual analogue scale is one possible way to gauge satisfaction (VAS). They have verified the VAS for satisfaction in this study. In this prospective investigation, 147 patients were examined (153 hips). The Harris hip score, the pain VAS at rest and during exercise, the Oxford hip score, the Short Form 36, and the Western Ontario McMaster Universities Osteoarthritis Index were used in the Spearman correlation test to determine the construct validity. The intra-class coefficient was used to assess dependability. According to the Pearson correlation test, correlations ranged from 0.40 to 0.80 in the result. Oxford hip score and the satisfaction VAS revealed a strong association, which may indicate that pain is one of the most significant variables in patient satisfaction. 0.95 was the intra-class coefficient. In the end, it was shown that the satisfaction VAS and the present subjective and objective scoring systems have a moderate to significant degree of association. Excellent test-retest reliability is indicated by the intraclass coefficient of 0.95. An easy tool to measure patient satisfaction following total hip replacement is the VAS satisfaction scale. This study has demonstrated the validity and reliability of the satisfaction VAS.

Ezzati A. at el (2014)[11]: have gone through the most used tools for measuring stress-the Perceived Stress Scale, which has three different variants(PSS-14, PSS-10, and PSS-4). Their primary objective was to validate this instrument in a sample of senior persons without dementia in order to facilitate research on the effects of stress on health. The PSS-14 questionnaire and additional neuropsychological tests were completed by a total of 768 non-demented persons over the age of 70. All PSS versions' underlying factor structures were discovered using exploratory factor analysis (EFA), and the construct validity of the factors was examined using confirmatory factor analysis (CFA). Using Cronbach's alpha, the scales' internal consistency reliability was analysed, and concurrent validity was assessed by looking at how the PSS related to variables including age, gender, depression, anxiety, and the Positive and Negative Affect Schedule (PANAS). The outcome demonstrates that a two-factor model provided the best match for the PSS 14- and 10-item versions. With the exception of item 12, all PSS-14 items had loadings greater than 0.4 for one of the two criteria. They have therefore tested a 13-item PSS version in addition to 10- and 4-item subsets that correspond to PSS-10 and PSS-4. Internal consistency coefficients for the PSS-13 and PSS-10 full scales, but not for the PSS-4, were satisfactory. Compared to men, women reported feeling more stressed. Higher total PSS scores were associated with lower levels of positive affect and higher levels of anxiety, depressive, and other negative emotions.

Cai W. et al (2018)[12]: have performed on sample of military people in this study to examine the characteristics of the Chinese version of the Anxiety Sensitivity Index-3 (ASI-3). The Chinese version of the ASI-3 was given to 3,077 valid individuals from nine military units, ranging in age from 16 to 36 (M = 22.35, SD = 3.57), using non-probabilistic sampling. The construct validity was evaluated using the State-Trait Anxiety Inventory (STAI) and the Depression Anxiety Stress Scales–21 (DASS-21). To compare the variations in the participants' positions and services, a one-way analysis of variance was done. The three-factor model of the original ASI-3 was found to have high convergent validity, and the confirmatory factor analysis indices were found to meet the standard values, supporting the hypothesis. On the other hand, the correlation between the ASI-3 and TAI was found to be relatively low, indicating that there was an empirical distinction between anxiety sensitivity and trait anxiety. The results of the

confirmatory factor analysis revealed that the indices satisfied the criteria, with the total scale's Cronbach's coefficient standing at 0.926 and the three subscales' values at 0.828–0.841; There were significant discoveries in the ASI and subscales among the five services and four positions, with sensitivity and specificity levels of 71.1% and 76.7%, respectively, at a cut-off score of 16, where the total of sensitivity and specificity becomes the maximum. This study adds to the body of evidence demonstrating the validity and reliability of the Chinese translation of the ANSI-3, which can be used to evaluate anxiety sensitivity in military personnel. The three element approach should be repeated across a few different special forces, and the items and constructions might be tweaked based on Chinese culture, they have advised scholars and practitioners.

4) **Partially edentulism**

McGarry TJ. at el (2002)[16]: Based on diagnostic results, the American College of Prosthodontists (ACP) has created a classification system for partial edentulism. This system of classification was comparable to the comprehensive edentulism classification system that the ACP had previously created. These recommendations were made to aid professionals in selecting the best courses of action for their patients. Class I represents an uncomplicated clinical state, and Class IV represents a complex clinical situation, making up the four classifications of partial edentulism that were defined. Specific diagnostic standards were used to distinguish each class. Dental experts who are diagnosing and treating partially edentulous patients should adopt this system. Improvements in intra-operator consistency, professional communication, insurance reimbursement commensurate with complexity of care, an improved tool for dental school admission clinics, standardised criteria for outcomes assessment and research, improved diagnostic consistency, and a streamlined referral decision aid are some potential advantages of the system.

5) **Salivary stress biomarkers and salivary alpha amylase (sAA)**

Speirs RL. et al (1974)[17]: have studied the effects of sympathetic activity and isoprenaline on amylase production from the human parotid gland. Using mild ascorbic acid applied to the tongue, human parotid saliva was generated and collected for this study. About 0.5 to 1.0 ml/min flow rates were attained. This baseline secretory activity was overlaid by sympathetic activation of the glands brought on by abrupt body cooling and inhalation or ingestion of isoprenaline. The salivary amylase concentration

significantly increased in the four study individuals in response to sympathetic stimulation. Increases in heart rate, blood pressure, and salivary flow rates were also present along with these alterations. Even while it was discovered that amylase activity rose marginally with higher flow rates, the significant increases in amylase concentration seen during sympathetic discharge could not be explained by variations in flow rate. The concentration of amylase in saliva triggered by acid was decreased when propranolol, a P-adrenergic blocker, was consumed.

Asking B. et al (1985)[18]: have examined the amylase secretion in the rat parotid gland. The simultaneous activation of both sympathetic and parasympathetic nerves and their separate stimulation were contrasted. When the two nerves were stimulated simultaneously, there was a significantly increased amylase output compared to when they were stimulated individually. Since parasympathetic activity was present in the background, it appeared that just 3-adrenoceptors were responsible for the sympathetic amylase secretion. Salivary flow was shown to be lowered when frequency (3–10 Hz) had been employed, and amylase output was also decreased. A-adrenoceptor blocking was used to mitigate this impact. It was shown that isoprenaline injections intravenously during a parasympathetic activity caused an increased amylase secretion similarly to sympathetic nerve stimulation.

Chatterton RT. et al (1997)[19]: have observed the purpose of this research was to look into how young males who were about to participate in a possibly fatal activity's hormonal and psychological reactions related to one another. To participate in their first skydiving leap, subjects were enlisted. Comparing ratings from the same participants taken 3-5 days prior to the leap with scores taken at 0800 h on the morning of the jump revealed no significant increase in anxiety scores. However, a psychological tool for assessing events showed a considerably higher intensity, and salivary amylase response, a measure of sympathetic nervous system activity, was higher than expected. Salivary cortisol and testosterone levels were considerably lower on the day of the jump than self-control and control result calculated during the same period. LH in plasma, on the other hand, was not suppressed. Just prior to the leap, the measurements of stress and anxiety as well as the evaluation of the events all reached high points. Serum cortisol, PRL, and GH all considerably enhanced after an increase in psychological measures, reaching peaks prior to or immediately post landing, and then noticeably

decreased over the course of the following hour, with the exception of testosterone, which stayed low. In the first 15 minutes after landing, anxiety and subjective stress ratings decreased to self-control levels, while the rating of events scale remained substantially high. In conclusion, reported anxiety related to a purely psychological stressor was repressed only a few hrs before the incident, but this inhibition was anticipated with an enhancement in sympathetic nervous system activity and a suppression of plasma cortisol. The actual incident was linked to reversing the cortisol fall; other stress-related chemicals increased, but salivary testosterone stayed stable.

Skosnik PD. et al (2000)[20]: have observed that two of the most salient physiological responses to stress are increased norepinephrine (NE) and cortisol (CORT) activities. It is not yet apparent how these neurochemical processes impact cognition, particularly attention. They looked at how little psychological stress affected their subjects' ability to pay attention selectively as measured by the negative priming NP. In order to examine the connection between the stress response and attentional inhibition, salivary measurements of the stress hormone CORT and a-amylase were performed. Healthy participants (N=20) participated in an attention task before spending 15 minutes playing a stressful video game before returning to the focus test. Prior to the experiment starting, 1 minute after the video game stressor, and 20 minutes later, baseline saliva samples were taken. After playing the video game, the subjects' NP and response time (RT) significantly decreased. A-amylase levels also markedly raised following the stressor, confirming NE's contribution to the acute stress response. Although CORT levels did not change following stress, CORT substantially linked with both NP and RT scores. These findings suggest that even minor psychological stress can have a major impact on attentional functions. It appears plausible that brain systems that regulate the fight-or-flight response tightly regulate attentional activities in light of the rise in a-amylase and the association between attention and CORT after stress.

Takai N. et al (2004)[21]: have observed that the non-invasive nature of saliva collection makes numerous samples simple and stress-free. Young people' salivary cortisol and amylase levels have been tested for their impacts, and the characteristics of these parameters have been compared. To determine their propensity for experiencing personal anxiety, the participants completed the STAI's trait version. For 15 minutes, a video of a corneal transplant procedure was used as a stressor. As a calming activity, a

movie of natural beauty was also employed. Unstimulated total saliva was extracted each three minutes throughout the entire session. The results show that shortly after the stressed video viewing began, the amylase level significantly jumped, and that it then abruptly returned to its pre-stress level just before the viewing came to a conclusion. Similar to how the level of amylase had grown, the level of cortisol had also gone up. Cortisol had a longer latency period before reaching its peak level than amylase. The leftover impact was seen in cortisol but not in the amylase response. Additionally, watching calming videos drastically reduced the amylase level while having little effect on the cortisol level. It is found that salivary amylase is a more accurate measure of stress than cortisol since it increased more considerably and responded more quickly to psychological stressors. Additionally, it has been hypothesised that the enzyme acts as a calming or relaxing index.

Gordis EB. et al (2006)[22]: have used Cortisol (C) and a-amylase (A-A), a proximate indicator of sympathetic nervous system (SNS) activity, for the samples. Adolescents' hostile behaviour was reported by parents, guardians, and other adults. Salivary A-A and C both increased as a result of the TSST, with a peak reaction for A-A occurring right away and for C 10 minutes later. Area under the curve was used to determine A-A and C stress reactivity (AUC). Adolescent violence as reported by parents varied by 7% due to asymmetrical C and A-A reactivity. Higher aggression evaluations were correlated with lower C reactivity at lower A-A reactivity levels, whereas C reactivity was unrelated to aggression at high A-A reactivity levels.

Proctor GB. et al (2007)[23]: have declared that "oral homeostasis is dependent upon saliva and its content of proteins. Reflex salivary flow occurs at a low 'resting' rate and for short periods of the day more intense taste or chewing stimuli evoke up to ten-fold increases in salivation. The secretion of salivary fluid and proteins is controlled by autonomic nerves". Acetylcholine is produced by cholinergic parasympathetic neurons, which feed all salivary glands. The majority of salivary glands also get variable degrees of sympathetic nerve innervation. Vasointestinal peptide, which is produced by the parasympathetic nervous system, aids in the reflex vasodilatation that occurs with secretion. It is believed that salivary protein production in reaction to autonomimetics is mediated by soluble, calcium-activated neuronal nitric oxide.Salivary glands need aquaporin 5 to produce fluid, however it's not known how much cholinomimetics can

actually boost aquaporin 5 expression on apical acinar cell membranes. It is yet unknown how salivary cells and neurons interact during salivary gland regeneration or growth.

Rashkova MR. et al (2012)[24]: have stated that salivary -amylase is a useful biomarker that may be used to assess human psychobiological and social behavioural processes. Its research provides opportunity for the creation of novel hypotheses about how biological and social processes interact and what impact they have on behaviour and health. Spielberger test scenario anxiety levels and salivary -amylase levels were measured twice in 30 individuals: once before the examination, under stressful conditions, and once a month afterwards, under less stressful conditions. Salimetrics LLC - USA's kinetic reaction kit was used to quantify salivary -amylase. The average level of salivary -amylase during the initial measurement was 156.0 93.33 U/ml, according to the results. The levels were two times lower during the second assessment, when there was no high stress present, and the changes were statistically significant (P 0.001). Between the amounts of salivary -amylase in the two assays, they discovered a statistically significant association (P 0.01). (P 0.01) The correlation coefficient was r = 0.472. It was recently determined that the modified State-Trait Anxiety Inventory score (STAI) developed by Spielberger was suitable for assessing stress-related anxiety in young people. Salivary -amylase can be employed as a biomarker for an unbiased assessment of a person's psychosomatic condition under a stressful situation. Salivary -amylase is a good example of an objective indicator that can be used in conjunction with psychological testing to assess a person's mental state under a stressful situation. Similar studies may be performed to evaluate patient behaviour during dental treatment, which is often thought to be stressful for patients.

Soeda R et al (2012)[25]: have selected the purpose of this study was to examine the effects of chewing force on salivary stress markers, including secretory immunoglobulin A secretion rate, salivary cortisol level, and alpha-amylase activity, which are indicators of mental stress. Twenty healthy men made up the participants. To assess stress markers, the first set of saliva samples (S1) was taken right after a 20-min break. The second set of saliva samples (S2) were taken after the subjects completed 20 minutes of arithmetic calculations as stress loading. The third set of saliva samples (S3) were then taken from each participant after they had chewed a piece of flavourless

gum for 10 minutes. The fourth batch of saliva samples (S4) was taken after a 20-minute break. They have assigned weak, ingrained, and strong chewing forces. Calculated were the rates at which stress markers changed between S2 and S3 and S2 and S4. Between S2 and S3, there was a discernible variation in the rate at which cortisol levels changed. Strong chewing reduced cortisol levels more than weak chewing. Between the three chewing forces, no discernible variations in s-IgA secretion rate or amylase activity change rate were found. The findings imply that variations in chewing power affect the levels of salivary cortisol for the three stress markers and that a strong chewing force reduces mental stress more than a weak one.

Schumacher S. et al (2013)[14]: have observed that in recent years, there has been an increase in interest in employing salivary alpha-amylase (sAA) as a non-invasive, substitute marker for sympathetic activity. Additionally, they noted that numerous research using stress protocols have shown that sAA is extremely sensitive to alterations brought on by stress (in healthy subjects). Additionally, it was proposed that sAA may also function as a marker for autonomic nervous system (ANS) pathological dysregulation in patients exhibiting psychopathology. Since then, sAA in individuals with mental illnesses has been the subject of a modest but developing literature. This review's objective was to provide a summary of early discoveries in this area of study. The findings of 15 investigations are thoroughly examined, along with the implications for future study. Changes in sAA, showing adrenergic dysregulation, might be demonstrated in psychopathology, notably in anxiety-related disorders, despite the fact that the number of research and the samples analysed were rather limited. The studies covered in this review, however, provided the first proof that using sAA as a sign of ANS dysregulation in mental disorders is promising.

Petrakova L. et al (2015)[10]: have reviewed many articles and stated that Plasma noradrenaline (NA) concentrations and salivary alpha-amylase activity (sAA) are frequently used as substitute indicators of stress. Moreover, reliability and generality of this interaction remain unclear. They used the Trier Social Stress Test (TSST). With no discernible variations between males and females, the TSST dramatically raised plasma levels of sAA and NA. However, neither group's sAA before, during, or after stress exposure substantially differed when subjects were categorised into low versus high responders based on their NA responses. These findings imply that levels of Plasma

noradrenaline and salivary alpha-amylase indicate acute psychological stress; yet, they appeared to rise independently of one another.

Klaus K. et al (2018)[15]: have gone through many research articles and written that, "although sleep is linked to physiological stress systems like the autonomic nervous system (ANS), research is still limited regarding night-and-day interactions between nocturnal sleep characteristics, stress, and diurnal parameters of salivary alpha-amylase (sAA) as a surrogate marker of ANS activity". In this study, fifty healthy university students underwent ecological momentary assessments over two five-day periods in low stress (at the start of the semester) and high stress (during the study) settings (final examination preparation). Six times a day, participants collected saliva and immediately after waking up, they reported on their previous night's sleep (quality, latency, duration, and interruptions). A sub-sample of participants also wore actigraphs to track their "time in bed." In contrast to earlier theories, under high stress, but not under low stress, poor sleep predicted lower sAA awakening values, more diminished awakening reactions, and steeper diurnal slopes the following day. The sAA values from one day to the next did not predict the sleep characteristics of the next night. The sAA profile appears to be a sign of more sustained stress-induced ANS dysregulation rather than being responsive to fluctuations in sleep that occur on a daily basis.

Rajagopal S. et al (2020)[26]: have observed that mastication is a stress-relieving activity in humans, according to clinical investigations. Lack of teeth affects chewing, which increases psychological stress in people and manifests in numerous physiologic changes to the body. Biomarkers for psychologic stress that are quantified serve as indicators of underlying physical illness. There is less research on how alpha amylase stress biomarkers function in partially edentulous clinical settings. This clinical study's objective was to compare sAA levels in partially dentate patients pre and post treatment of fixed dental prosthesis. For this investigation, 40 patients with missing mandibular first molars and partial dentition were chosen. Participants' levels of stress and anxiety were assessed using two questionnaires: The State Trait Anxiety Inventory (STAI) and the Perceived Stress Scale (PSS). Metal ceramic fixed dental prostheses were used to treat the selected subjects (FDP). To gauge patient satisfaction, a visual analogue scale (VAS) was employed. Unstimulated saliva samples were taken preoperatively, three months later, and six months after the FDP was implanted. The sAA level was

estimated. The paired sample t-test (=.05) was used for statistical analysis of the mean and standard deviation (SD) data. According to the findings, before FDP restoration, the salivary alpha amylase level was at its maximum, with a mean of 36.73 M/min/mg ptn. The enzyme levels dropped to 16.62 M/min/mg ptn in the third month following prosthesis placement, and the lowest value of 8.58 M/min/mg ptn was found in the sixth month (P 0.05). It is concluded that after tooth replacement with FDP, the salivary alpha amylase stress indicator decreased.

METHODOLOGY

Data collection

Patients with missing maxillary or mandibular teeth who are partially edentulous will be chosen, and those who agreed to participate in the study will be enrolled.

Based on clear inclusion and exclusion criteria, patients will be chosen.

- **Inclusion Criteria**

  The inclusion criteria for the trial will be stable physical and mental health, partial edentulous status lasting no longer than six months, and absence of spacing, misalignment, or temporomandibular joint problem.

- **Exclusion Criteria**

  The study will exclude subjects with eating disorders like anorexia nervosa and bulimia nervosa as well as those with psychotic disorders, systemic illnesses, pregnant women, individuals having habit of smoking, individuals receiving steroid therapy within the past 3 months, autoimmune diseases, old denture wearers, complete edentulism states, and illness of the salivary glands.[16]

To check the participants' anxiety and psychological stress, the State Trait Anxiety Inventory (STAI) and the PSS questionnaire (Sheldon Cohen) will be given to chosen subjects. The study's participants will be chosen from those who scored low on the questionnaire's measures of stress and anxiety. Participants that will be enrolled, will have their missing tooth restored

using a standard full veneer ceramo-metal FDP. For fixed dental prostheses, standardised treatment guidelines will be followed.

Before the treatment, one month after treatment, and two months afterwards, salivary samples will be taken. The normal procedure for obtaining salivary samples will be followed.[24] Morning appointments between the hours of 10.00 am to 2.00 pm will be booked 45 minutes after brushing teeth. Prior to the session, participants will be instructed not to engage in strenuous physical exercise and refrain from consuming any food or beverages. The day before sample collection, no dental treatment should be done. Additionally, they will be forbidden from consuming any drugs, alcohol, or caffeine 12 hours before their visit. They will be told to use mouthwash to remove any food leftovers from their mouths on the day of the samples collection.

Salivary samples will be taken 15 minutes after the waiting time. Using a passive drool technique, the participant's unstimulated saliva will be collected in a sterile plastic 5 mL cryovial. In order to drive the saliva into the container, the patient will have to gently collect saliva under the tongue and lean their head downward. The obtained sample will be centrifuged for 10 minutes at 3000 g. This will create a supernatant solution, that will then be transferred using a pipette to another cryovial and kept at -80 °C. For the transportation of samples to the testing laboratory for the determination of sAA, an insulator freezer box will be employed.[27]

Data analysis

Alpha-amylase levels for each individual will be recorded at each time point. To compare mean values over various intervals, a paired-sample t-test will be used. The difference in mean values between the sexes will be assessed using an independent-sample t-test. Using (SPSS v16; SPSS Inc), Chicago, IL, USA, the data will be examined.

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